



## A new species of *Bolitoglossa* (Caudata, Plethodontidae) from the continental divide of western Panama

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### Abstract

We describe the new salamander species *Bolitoglossa jugivagans* from the Atlantic slopes of the Fortuna depression in western Panama on the basis of morphological and molecular data. Based on mtDNA data, the new species seems to be closely related to *B. aureogularis* and *B. robinsoni*, with which it forms a subclade within the subgenus *Eladinea*.

**Key words:** *Bolitoglossa jugivagans* sp. nov., *B. robinsoni* clade, La Fortuna Forest Reserve, Protected Forest Palo Seco, DNA barcoding, *Eladinea*, Serranía de Talamanca

### Resumen

Describimos la nueva especie de salamandra *Bolitoglossa jugivagans* de la vertiente atlántica del rebajamiento de Fortuna en Panamá occidental a base de datos morfológicos y moleculares. Basado en datos de ADN mitocondrial, *B. robinsoni* y *B. aureogularis* parecen ser las especies más cercanamente relacionadas con la nueva especie, las tres formando un subclado dentro del subgénero *Eladinea*.

**Palabras claves:** *Bolitoglossa jugivagans* sp. nov., clado *B. robinsoni*, Reserva Forestal La Fortuna, Bosque Protector Palo Seco, código de barras de ADN, *Eladinea*, Serranía de Talamanca

### Introduction

The Cordillera Central of Costa Rica and Panama is a unique realm of salamander diversification on a worldwide scale. While there are only the three genera *Bolitoglossa*, *Nototriton*, and *Oedipina* of the sole family Plethodontidae in these mountains, the species diversity is extraordinary high (Bolaños and Wake 2009). Among Panamanian amphibians, *Bolitoglossa* comprises the second highest species count after the frog genus *Craugastor* (Jaramillo *et al.* 2010; Hertz *et al.* 2012). Above all it is, with currently 123 described species (AmphibiaWeb 2012), the largest genus in the order Caudata worldwide. On the basis of mtDNA data Parra-Olea *et al.* (2004) recognized several subgenera in *Bolitoglossa*, two of which are found in Panama. According to this classification, most Panamanian species belong to the subgenus *Eladinea*, with *B. lignicolor* (Peters) being the only representative of the subgenus *Bolitoglossa*. In 2009, we collected a single individual of *Bolitoglossa* (*Eladinea*) near the Fortuna depression, almost on the continental divide. This specimen is morphologically different from all described species, but since we had only a single specimen we decided to postpone the formal description until additional specimens were available. However, despite of intensive field work in that area in the subsequent year, no other specimen could be recovered. Recent fieldwork by Boza-Oviedo *et al.* (2012) in the little investigated high Talamancan Mountains in eastern Costa Rica near the border with Panama produced five new species of salamanders. Furthermore, they presented molecular data for several of poorly known salamanders from the central

mountain ranges of Costa Rica. Based on molecular evidence they discovered a formerly unknown clade within the subgenus *Eladinea*, hereinafter referred to as the *B. robinsoni* clade, which at that time contained the two species *B. robinsoni* Bolaños and Wake and *B. aureogularis* Boza-Oviedo, Rovito, Chaves, García-Rodríguez, Artavia, Bolaños, and Wake. After the availability of this paper, we compared a 16S rRNA gene sequence of our unidentified salamander from Panama to the sequences provided by Boza-Oviedo *et al.* (2012). We found it to be the henceforth third member of the *B. robinsoni* clade, however, with still large genetic distances to its closest relatives. This additional evidence obliges us to describe it as a new species.

## Material and methods

Preparation and preservation of the holotype follows Köhler (2001). A small piece of muscle from the lateral tail has been removed as tissue sample, stored in 98% undenatured ethanol and deposited in the tissue collection of the Senckenberg Forschungsinstitut und Naturmuseum Frankfurt, Germany (SMF). Abbreviations for museum collections for comparative voucher specimens follow those of Sabaj Pérez (2010). Geographic coordinates and altitude above sea level of the type locality were recorded with a handheld Garmin MAP 60 CSx GPS receiver. The georeference is in the geographical coordinate system and WGS 1984 datum, given in decimal degrees rounded to the fourth decimal place. Elevations are rounded to the nearest tenth. The map was created using ArcGIS 10 (ESRI) with GIS layers from the Smithsonian Tropical Research Institute (STRI) map server (<http://mapserver.stri.si.edu/>). We conducted automatized temperature and relative humidity measurements at the type locality of the new salamander once per hour using an iButton® datalogger (Maxim Integrated<sub>TM</sub>). Additionally, we measured precipitation for the time of our stay using a Conrad Electronics rain gauge (KW-9015). Average climate data are derived from the WorldClim database (Hijmans *et al.* 2005).

The morphological characters of the holotype follow the methodology of Boza-Oviedo *et al.* (2012) and were made with a dial precision caliper under a dissecting microscope (Leica MZ 12) rounded to the nearest 0.1 mm: snout-vent length (standard length), from tip of snout to posterior end of vent (SVL), tail length from posterior end of vent to tip of tail (TL), gular fold to tip of snout (SG), head width at greatest width of head (HW), head depth at posterior angle of jaw (HD), eyelid length (EL), eyelid width (EW), anterior margin of orbit to tip of snout (ES), horizontal eye diameter (ED), intercanthal distance (IC), interorbital distance between eyelids (IO), tip of snout to forelimb (SF), internarial distance (IN), snout projection (SP), shoulder width (SW), snout to anterior angle of vent (SAV), axilla-groin distance (AX), hind limb length from groin to tip of longest digit (HLL), forelimb length from axilla to tip of longest digit (FLL), hand width at widest extent (HAW), hind foot width at widest extent (FW), length of third toe (T3), length of fifth toe (T5). Premaxillary teeth (PMT), maxillary teeth (MT), and vomerine teeth (VT) were counted under a dissecting microscope. MT and VT are provided for left and right sides, respectively. We further counted costal folds between the adpressed limbs of the straightened specimen (limb interval, LI) as a measure of relative limb length. The capitalized colors and color codes (the latter in parentheses) used in the description of the coloration in life are those of Smithe (1975–1981). For a preliminary designation of the collected specimen we employed the keys to the genus *Bolitoglossa* by Savage (2002) and Köhler (2011). Morphological characters and tooth counts for comparison were taken from the original species descriptions by Bolaños *et al.* (1987), Bolaños and Wake (2009), Wake *et al.* (2007), and Boza-Oviedo *et al.* (2012), and tooth counts for *B. subpalmata* from Ehmcke and Clemen (2000). Information on the osteology has been derived from radiographs of the holotype.

A fragment of the mitochondrial 16S rRNA gene of the holotype was sequenced for comparison with a set of sequences available on GenBank. DNA was extracted from muscle tissue following the protocol of Ivanova *et al.* (2006). The mitochondrial 16S rRNA gene was amplified using a Mastercycler pro S (Eppendorf, Hamburg, Germany) performing an initial denaturation for 1 min at 94° C followed by 35 steps with denaturation for 0.25 min at 94° C, hybridization for 0.75 min at 45° C, and elongation for 1.5 min at 72° C, final elongation proceeded for 7 min at 72° C. Reaction mix contained 1 µL DNA template, 2.5 µL Reaction Buffer x10 (PeqGold), 4 µL 2.5 mM dNTPs, 0.4 µL (containing 2.5 units) Taq Polymerase (PeqLab), 14.1 µL H<sub>2</sub>O, 1 µL 25 mM MgCl<sub>2</sub>, and 1 µL (containing 10 pmol) of each of the universal primers 16SA-L and 16SB-H (Palumbi *et al.* 1991). A list of specimens included in the genetic analysis with corresponding GenBank accession numbers is in the appendix. Sequences were aligned with ClustalW (Larkin *et al.* 2007) using the default settings in Geneious (Drummond *et*

al. 2010). *Bolitoglossa lignicolor* was used as an outgroup. The manually refined final alignment was trimmed to the length of the shortest sequence and then consists of 399 positions of which, excluding the outgroup, 62 were variable and 43 parsimony-informative. Using MEGA5 (Tamura *et al.* 2011), we computed uncorrected pairwise genetic distances, determined the Tamura 3-parameter model (Tamura 1992) as the best-fitting substitution model, and conducted Maximum Likelihood analysis (with 1000 bootstrap replicates). We further ran a Bayesian phylogenetic analysis in MrBayes 3.1.2 (Huelsenbeck and Ronquist 2001). We used JModeltest 0.0.1 (Posada 2008) under the Akaike Information Criterion (AIC) to select the substitution model for the Bayesian analysis. The analysis was run for 2,000,000 generations with four default chains, sampled every 100 generations. We then discarded 5 % of these generations as burn-in. Convergence of runs was assessed using the AWTY graphical analysis (Nylander *et al.* 2007). Using TCSv1.21 (Clement *et al.* 2000), we conducted a statistical parsimony network analysis of the *Bolitoglossa robinsoni* clade with gaps considered as a fifth character state. In order to connect all haplotypes we set the connection limit to 15 steps. Figure 3 and 8 were created using GraphPad Prism. All Figures have been graphically improved and combined using Adobe CS3.

## Results

### *Bolitoglossa jugivagans* sp. nov.

Continental Divide Salamander

Figures 4–7

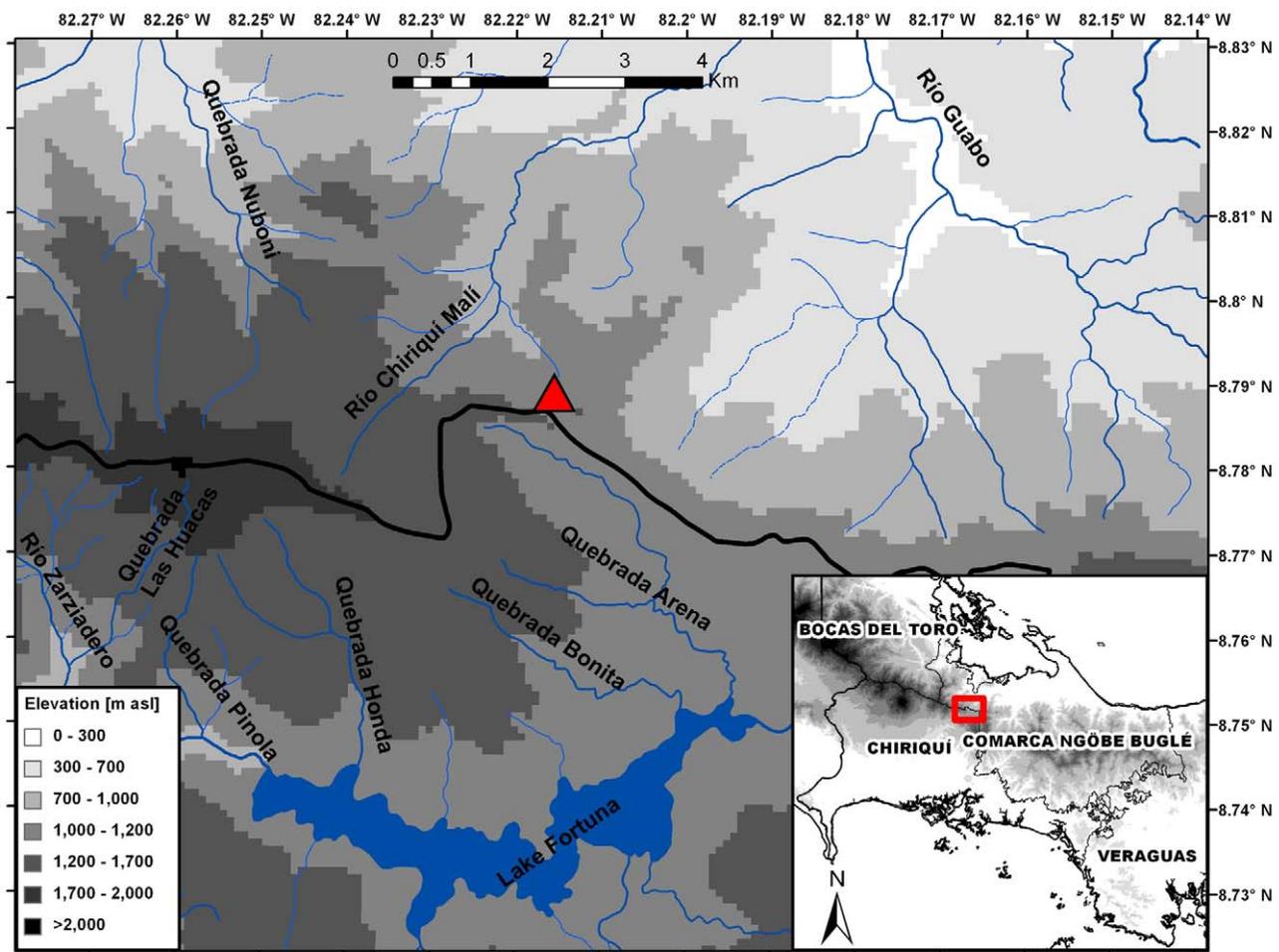
**Holotype.** SMF 94467 (original field number AH 314), an adult female from the headwaters of Río Chiriquí Malí, approximately 5 km N of the La Fortuna dam reservoir (8.7890°N, 82.2154°W, 1060 m), Bosque Protector Palo Seco, Comarca Ngöbe-Buglé (formerly province of Bocas del Toro), Panama (Fig. 1); collected by Andreas Hertz and Sebastian Lotzkat on 28 October 2009.

**Diagnosis.** We assign the new species to *Bolitoglossa* because it has fewer than 14 costal grooves and lacks a sublingual fold (Savage 2002), and to the subgenus *Eladinea* based on mtDNA sequence data. It differs from all described species by the following combination of characters: *Bolitoglossa jugivagans* is a small species with short legs and a long tail. The hands and feet are moderately webbed, with the two terminal phalanges of the longest digits free of webbing. The prehensile tail is considerably longer than the body. Both maxillary and vomerine tooth counts are comparatively high in relation to SVL.

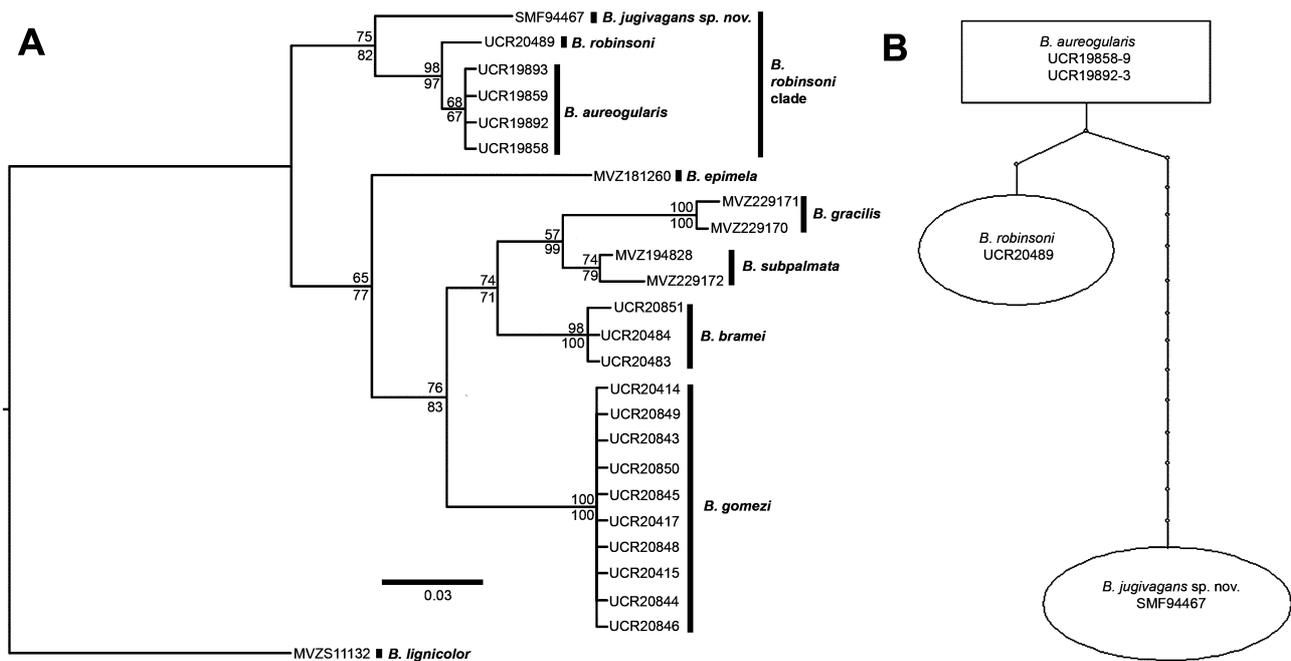
*Bolitoglossa jugivagans* is well differentiated from all described species of the subgenus *Eladinea* by 16S mtDNA distances (Fig. 2; Table 1). It is further distinguished morphologically by the following characters (condition for *B. jugivagans* in parentheses): It differs from *B. biseriata* Tanner, *B. bramei* Wake, Savage, and Hanken, *B. colonea* (Dunn), *B. lignicolor*, *B. minutula* Wake, Brame, and Duellman, *B. schizodactyla* Wake and Brame, and *B. epimela* Wake and Brame by its less extensively webbed hands and feet. All these species have fully webbed hands and feet, or at least less than two phalanges free of webbing (slightly more than two phalanges free of webbing). *Bolitoglossa jugivagans* is easily distinguishable by coloration (described below in coloration section) from the large, almost entirely black species *B. anthracina* Brame, Savage, Wake, and Hanken, *B. compacta* Wake, Brame, and Duellman, *B. copia* Wake, Hanken, and Ibañez, *B. magnifica* Hanken, Wake, and Savage, *B. nigrescens* (Taylor), *B. robusta* (Cope), and *B. sombra* Hanken, Wake, and Savage. It can be differentiated from other small salamanders with high tooth counts from the Talamanca Mountains as follows: *Bolitoglossa pygmaea* Bolaños and Wake is almost unpigmented and has a short tail that never exceeds SVL (*B. jugivagans* has a normal reddish-brown pigmentation and the tail is much longer than SVL). *Bolitoglossa gracilis* Bolaños and Robinson is bright yellowish to golden tan with dark streaks and spots (reddish-brown with light streaks), has longer legs with a limb interval of 3 to 3.5 (4) and fewer vomerine teeth VT: 10–22 (27; see also Fig. 3). *Bolitoglossa pesrubra* Taylor, with which the new species shares high vomerine and maxillary tooth counts, has longer legs with a limb interval of 0 to 3 folds (4) and a shorter tail TL/SVL $\approx$ 1 (TL/SVL=1.24). *Bolitoglossa subpalmata* (Boulenger) has fewer maxillary and vomerine teeth at the same size (Fig. 3) and a limb interval of 1 to 3 (4). *Bolitoglossa gomezi* Wake, Savage, and Hanken has a shorter tail TL/SVL: 1.15–1.19 in females (1.24), limb interval of 1.5–3 (4), and fewer maxillary and vomerine teeth at a comparable SVL (Fig. 3). *Bolitoglossa kamuk* Boza-Oviedo, Rovito, Chaves, García-Rodríguez, Artavia, Bolaños, and Wake has a dark ground coloration with golden flecking on the posterior

part of the body and the tail (reddish-brown with light streaks), as well as fewer maxillary and vomerine teeth MT: 34 (51), VT: 13–16 (27), and a shorter tail  $TL/SVL \approx 1$  ( $TL/SVL = 1.24$ ). *Bolitoglossa splendida* Boza-Oviedo, Rovito, Chaves, García-Rodríguez, Artavia, Bolaños, and Wake is peculiarly colored with a bright-red dorsal band and bright-yellow spots laterally and ventrolaterally on jet black ground coloration (reddish-brown with light streaks), has only 18 vomerine teeth at a SVL of 47.8 mm (27 at  $SVL = 31.2$  mm), has more webbing on feet that extends to between first and second phalangeal articulations of longest digits (two terminal phalanges free on the longest digits), and a broader head  $HW/SVL = 0.17$  ( $HW/SVL = 0.14$ ).

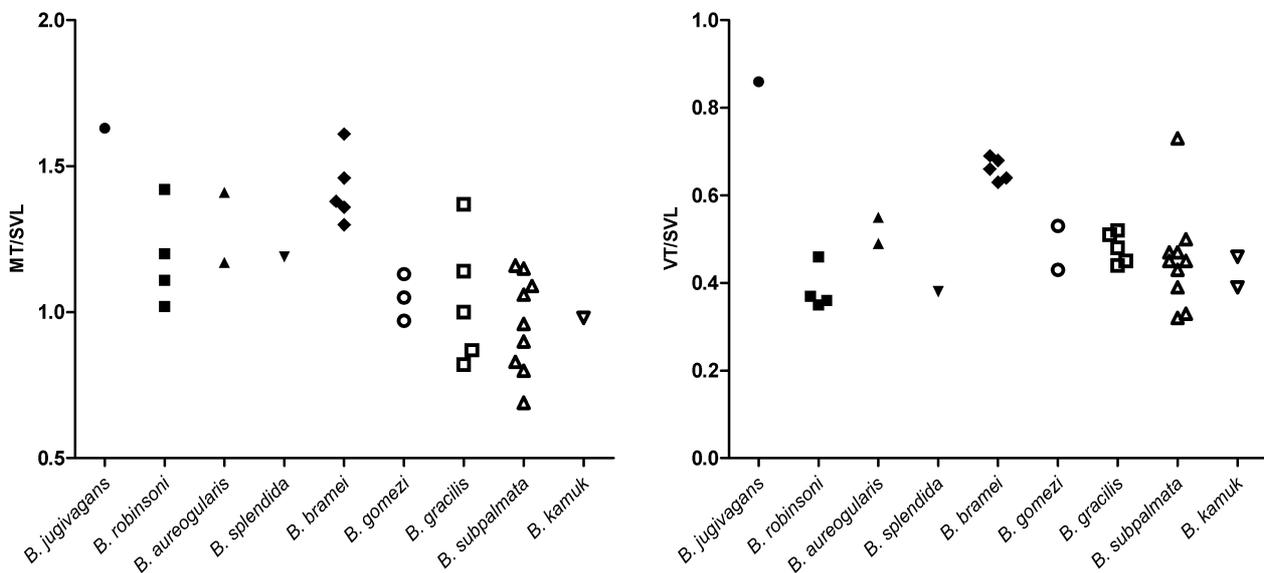
Based on molecular data, *Bolitoglossa jugivagans* is most closely related to *B. robinsoni* and *B. aureogularis*. Both differ from *B. jugivagans* by the following morphological characters (condition for *B. jugivagans* in parentheses): *Bolitoglossa robinsoni* is a large and stout species with a broad head  $HW/SVL = 0.16–0.17$  (0.14) and a tail that is shorter than the standard length  $TL/SVL \leq 1$  ( $TL/SVL = 1.24$ ). The second species *B. aureogularis* is superficially very similar to *B. jugivagans*, primarily in dorsal coloration. However, *B. aureogularis* has fewer maxillary and vomerine teeth at a comparable size (Fig. 3) and much shorter legs with a limb interval of 6.5 in females and 5 in males (4 in the female holotype of *B. jugivagans*). *Bolitoglossa aureogularis* differs most markedly from *B. jugivagans* in ventral coloration, having a bright yellow gular region and a prominent midventral dark stripe on dirty white ground (gular region mustard yellow; ventral coloration of body grey to brownish speckled with blue, no conspicuous dark midventral stripe).



**FIGURE 1.** Map showing the type locality (red triangle) of *Bolitoglossa jugivagans* on the banks of a feeder river of Río Chiriquí Malí. Further important watercourses are shown in blue. The black line represents the provincial border between Chiriquí in the south and the Comarca Ngöbe-Buglé in the north (cf. inset), and at the same time the border between the protected areas Reserva Forestal La Fortuna (south) and Bosque Protector Palo Seco (north), as well as the drainage divide between river systems flowing into the Pacific Ocean (south), and those flowing into the Atlantic Ocean (north). The elevation of the high point west of the type locality is Cerro Guayabo (approx. 2000 m asl).



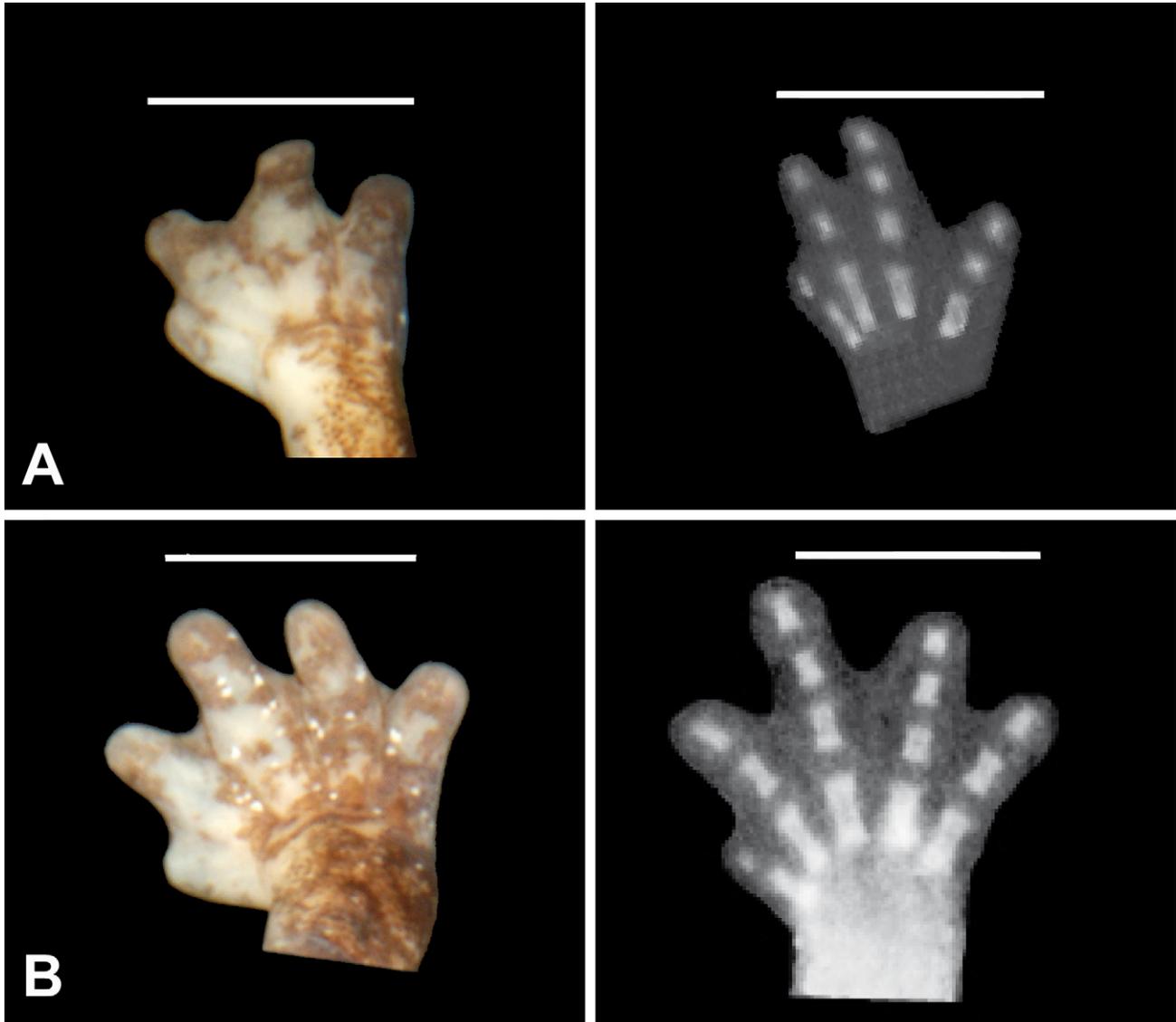
**FIGURE 2.** Results of the molecular analyses. (A) Phylogeny of 16S mtDNA data for *Bolitoglossa jugivagans* compared to selected specimens of *Bolitoglossa (Eladinea)* labeled with museum catalogue numbers, constructed using Bayesian analysis. Scale bar refers to number of substitutions per site. Maximum likelihood bootstrap values are shown above branches, Bayesian posterior probabilities (multiplied by 100) below branches. Taxonomic identities according to Boza-Oviedo *et al.* (2012). (B) Parsimony network with a connection limit of 15 steps; each node represents a unique haplotype separated from the next by one substitution step. Small circles represent unsampled haplotypes, the rectangle the probably ancestral haplotype.



**FIGURE 3.** Tooth counts in relation to SVL of *Bolitoglossa* species resembling each other morphologically (males and females are shown together; see Material and Methods for source of data). (A) Maxillary teeth (MT) divided by snout-vent length (SVL). (B) Vomerine teeth (VT) divided by snout-vent length (SVL).

**Description of the holotype.** This is a small and slender *Bolitoglossa*. The holotype is an adult female of 31.2 mm SVL; tail long and prehensile, TL/SVL is 1.24; head short (HL about 21 % of SVL) and narrow (HW slightly less than 14 % of SVL); hands and feet narrow (HAW = 7 %, HFV = 9 % of SVL), and limbs short (HLL about 20

% of SVL, limb interval 4 costal folds between adpressed limbs); webbing moderate with the two terminal phalanges free on the longest digits (Fig. 4); order of increasing length of digits I-IV-II-III in hands, and I-V-II-IV-III in feet; subterminal pads present on longest digits; small eyes that do only slightly protrude beyond lateral margins of head in ventral view; labial protuberances weakly developed; teeth moderate in size and numerous (PMT 4, MT 51, VT 27), maxillary teeth extending to about level of center of eye; prevomer clearly projecting from level of palate, bearing vomerine teeth in long, slightly arched, irregular series extending laterally well beyond outer margin of choanae.



**FIGURE 4.** Dorsal view and x-ray images of hands and feet of *Bolitoglossa jugivagans*. (A) Right hand in dorsal view with corresponding x-ray image to the right. (B) Right foot in dorsal top view with corresponding x-ray image to the right. Scale bar = 2 mm.

**Measurements (in mm), tooth counts and limb interval of the holotype.** SVL 31.2, TL 38.8, HW 4.3, SG 6.5, HD 2.5, EW 0.7, EL 1.5, ES 1.8, ED 1.3, IC 2.2, IO 1.6, SF 8.4, IN 1.0 SP 0.6, SW 3.6, SAV 28.7, AX 17.8, LI 4, FLL 4.8, HLL 6.2, HAW 2.2, FW 2.8, T3 1.3, T5 1.0, Numbers of teeth: PMT 4, MT 23/28, VT 12/15.

**Coloration of the holotype in life** (Fig. 5). Dorsal ground color Chestnut (32), interspersed with Fuscous (21) and Salmon Color (106) broken longitudinal lines. A Salmon Color (106) dorsolateral line running from eye to groin separates the middorsal coloration from the lateral and ventral ground coloration; ventral ground coloration Sulfur Yellow (157) flecked with gray at night and Fuscous (21) at daytime. Ventral surfaces speckled with Sky Blue (66). Gular region, hands, and feet Sulfur Yellow (157) suffused with Fuscous (21) on limbs.



**FIGURE 5.** Holotype of *Bolitoglossa jugivagans* in life. (A) Holotype at the moment of encounter at night. (B) Holotype the subsequent day. Note difference in lateral coloration.

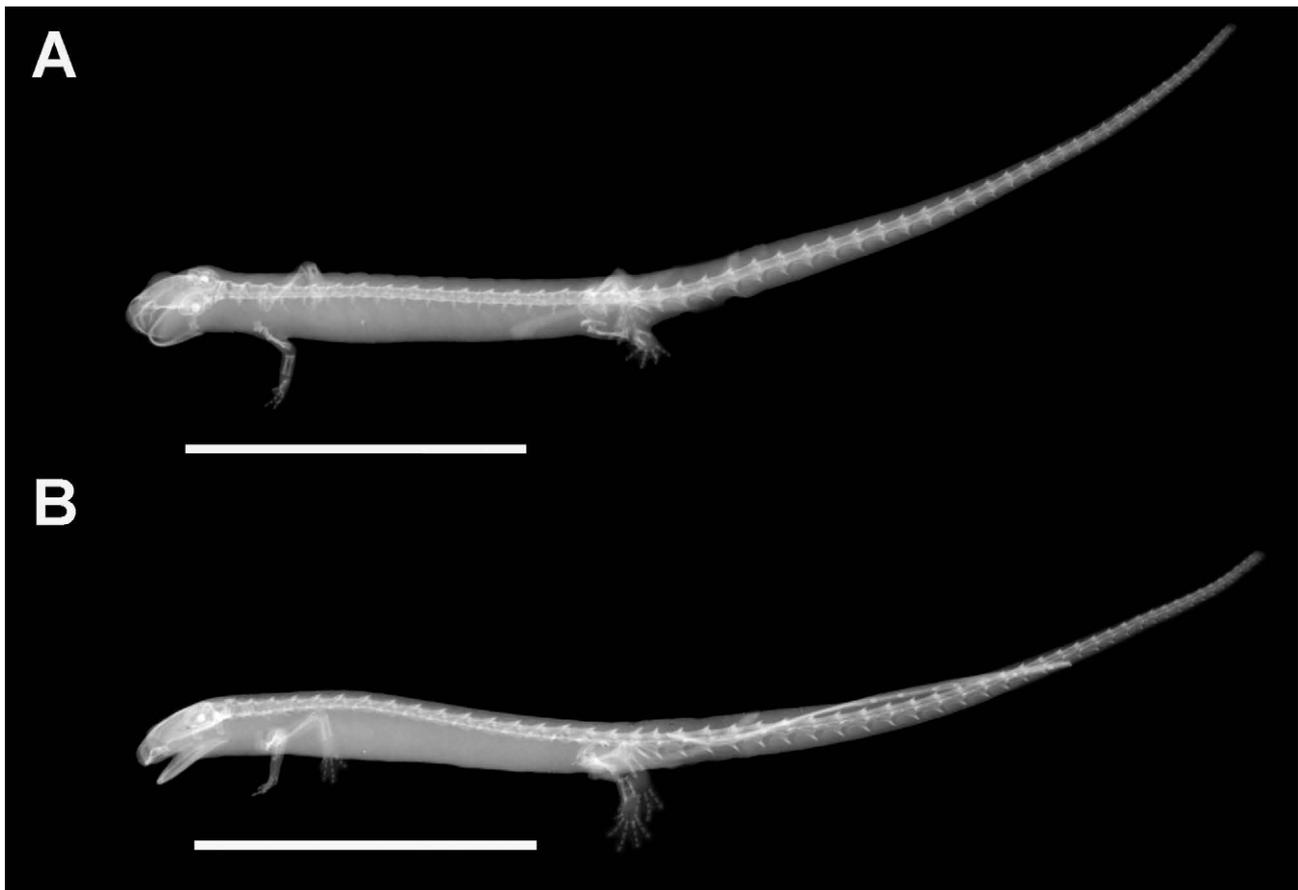
**TABLE 1.** Mean genetic distances in the 16S rRNA gene between *Bolitoglossa* species used in the phylogenetic analysis. Numbers above diagonal are for Tamura-3-parameter-distances and numbers below are uncorrected p-distances.

Species		<i>Bjug</i>	<i>Brob</i>	<i>Baureo</i>	<i>Bbra</i>	<i>Bgom</i>	<i>Bsubp</i>	<i>Bgraci</i>	<i>Bepim</i>	<i>Bligni</i>
<i>B. jugivagans</i>	<i>Bjug</i>	-	0.042	0.039	0.062	0.075	0.067	0.088	0.068	0.103
<i>B. robinsoni</i>	<i>Brob</i>	0.040	-	0.008	0.065	0.067	0.058	0.080	0.068	0.106
<i>B. aureogularis</i>	<i>Baureo</i>	0.038	0.008	-	0.062	0.064	0.056	0.077	0.065	0.103
<i>B. bramei</i>	<i>Bbra</i>	0.059	0.062	0.059	-	0.046	0.035	0.039	0.063	0.107
<i>B. gomezi</i>	<i>Bgom</i>	0.071	0.063	0.061	0.045	-	0.042	0.060	0.065	0.105
<i>B. subpalmata</i>	<i>Bsubp</i>	0.063	0.056	0.053	0.034	0.040	-	0.034	0.065	0.111
<i>B. gracilis</i>	<i>Bgraci</i>	0.082	0.075	0.072	0.038	0.057	0.033	-	0.078	0.125
<i>B. epimela</i>	<i>Bepim</i>	0.065	0.065	0.062	0.060	0.062	0.062	0.074	-	0.120
<i>B. lignicolor</i>	<i>Bligni</i>	0.095	0.097	0.095	0.098	0.097	0.102	0.114	0.109	-

**Coloration of the holotype in alcohol** (Fig. 6). After three years in ethanol (60 %), its coloration has not changed much. Yellowish colors have faded to cream-colored shades as well as the reddish tones in the brown dorsal coloration.



**FIGURE 6.** Holotype after three years in 60 % ethanol; (A) Dorsal view; (B) ventral view. Scale bar = 2 cm.



**FIGURE 7.** X-ray images of the entire holotype; (A) Ventrolateral view; (B) Lateral view. Scale bar = 2 cm.

**Osteology** (Fig. 7). The radiographs of the holotype show the typical osteological features of the genus *Bolitoglossa*: Head well ossified; vertebral column consists of one atlas, 14 trunk vertebrae, one sacral and two caudosacral vertebrae, and 35 caudal vertebrae; digits well developed on all limbs; slightly more than two distal phalanges of the longest fingers and toes free of webbing, phalangeal formulae of the hand 1–2–3–2, and the foot 1–2–3–3–2. Terminal phalanges of longer fingers and toes slightly expanded distally (Fig. 4).

**Habitat and natural history notes.** *Bolitoglossa jugivagans* is only known from the type locality, the upper section of the valley of a feeder stream of Río Chiriquí Malí, on the Caribbean slope of the Cordillera Central, not far from the continental divide in the Fortuna depression of the Cordillera Central of western Panama (Fig. 1). The site is easily reachable on a paved road that serves as a maintenance access way to the Trans-Panama Oil Pipeline. The riverbank next to the street is an old clearing, overgrown with grass, herbaceous plants, and bushes, while the opposite bank is predominantly covered by broadleaf, evergreen mature forest. Mean annual precipitation is approximately 3000 mm and mean annual temperature approximately 20.6° C, making the site assignable to the Premontane Wet Forest life zone according to the Holdridge (1967) classification.

The holotype was encountered on October 28, 2009 at 23:00 h active on the large leaf of an Araceae plant growing out of the long grass on the cleared side of the river. That day, it had been raining in the afternoon and evening; our pluviometer registered 44.5 mm of precipitation between 16:00 and 24:00 h. The automatically recorded temperature was 19.6° C between 18:00 and 23:00 h, and then gradually decreased to a minimum of 17.6° C at 06:00 h. Relative humidity was constantly high at close to 100%. *Bolitoglossa jugivagans* was the only salamander encountered at this site. Other amphibian species collected in the vicinity include *Craugastor fitzingeri* (Schmidt), *Diasporus* sp., *Duellmanohyla uranochroa* (Cope), *Espadarana prosoblepon* (Boettger), *Incilius coniferus* (Cope), *Pristimantis cruentus* (Peters), *P. pardalis* (Barbour), and *Smilisca phaeota* (Cope). Additionally this site is the type locality of the recently described snake species *Sibon noalamina* Lotzkat, Hertz, and Köhler. Lotzkat *et al.* (2012) give additional information on the reptile fauna of the surroundings.

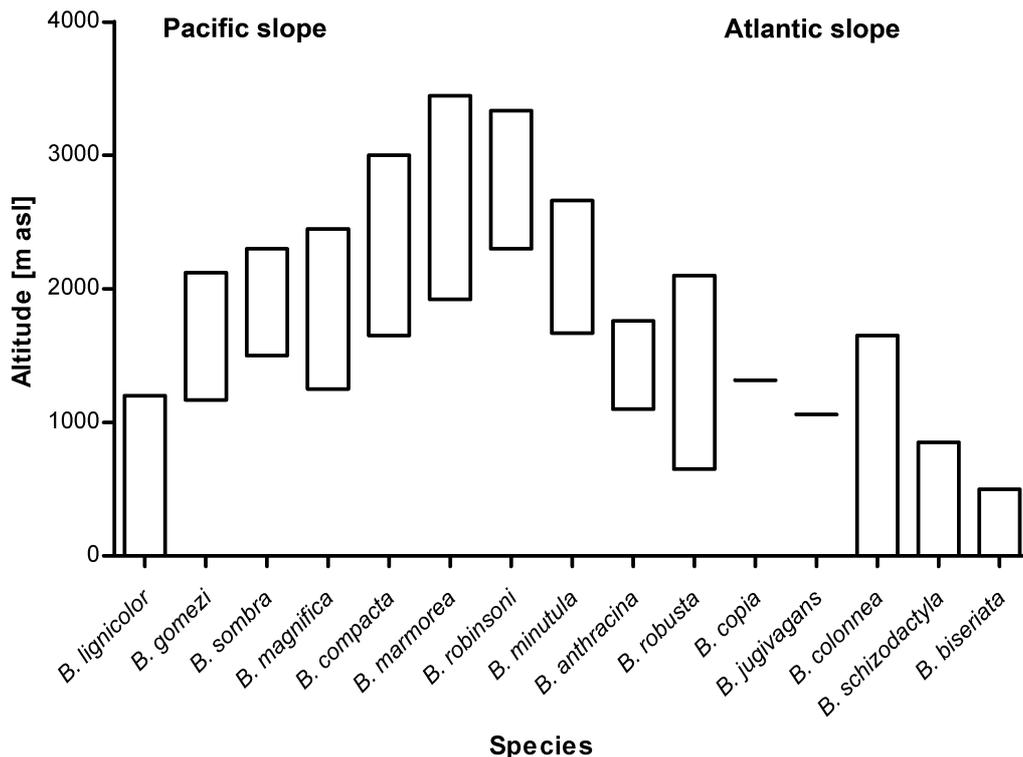
**Etymology.** The species name is a compound adjective derived from the locative of the Latin noun *jugum*

meaning “mountain ridge” and the present participle of the Latin verb *vagare* meaning “to wander”. The species epithet signifies “wandering on the mountain ridge” referring to the type locality next to the continental divide of the Cordillera Central.

## Discussion

The holotype and only known specimen has distinctly more maxillary and vomerine teeth in relation to its snout-vent length than any superficially similar species (Fig. 3). Regarding molecular data, the species is clearly separated from other species in the subclade *Eladinea*. The 16S rRNA gene has been demonstrated to be very suitable as a universal barcoding marker in amphibians (Vences *et al.* 2005). In this group a minimum threshold of 3 % uncorrected pairwise genetic divergence in the 16S rRNA gene to all other described species has been demonstrated to distinguish deep genealogical lineages, which might represent taxa on the species level if supported by additional species-specific differences (Vieites *et al.* 2009; Jansen *et al.* 2011). A p-distance of 3.8 % was calculated between *Bolitoglossa jugivagans* and its supposedly closest congener *B. aureogularis* (Table 1). Although the sample of available specimens in both species is very small, they are well differentiated morphologically, readily justifying the description of *B. jugivagans* as a new species. *Bolitoglossa aureogularis* has fewer maxillary and vomerine teeth for its size and is also generally larger. However, since we have only a single specimen of the new species, we avoided using SVL itself as a diagnostic character. Nonetheless, tooth counts and general appearance of the holotype imply that it is an adult specimen of female sex, the latter because cloacal folds are present. Besides tooth counts, the limb interval is a character that differentiates *B. aureogularis* from *B. jugivagans* with no overlap. In the text body of the original description of *B. aureogularis*, Boza-Oviedo *et al.* (2012) denote a limb interval of 3.5 folds, whereas Table 2 in the same work gives 7 folds for the female and 5 for the male. David Wake (pers. comm.) clarified that the correct measurements are those of Table 2, with the exception that the accurate limb interval of the female holotype is not 7 but rather 6.5, which was later reconfirmed by Gerardo Chaves (pers. comm.) who kindly examined the type material again. In many *Bolitoglossa* species, females tend to have relatively shorter legs than males (e.g., Brame & Wake 1963; Wake & Lynch 1982; Hanken *et al.* 2005; Rovito *et al.* 2012). Therefore, we expect males of *B. jugivagans* to have a limb interval of 2.5 to 3 folds between adpressed limbs. Concerning habitat preferences, *B. aureogularis* has been found at elevations between 1680 and 2100 m asl, thus considerably higher than *B. jugivagans*. However, we do not attribute much significance to that fact, as some *Bolitoglossa* species inhabit quite a large vertical range and the extension of the known vertical ranges along with the collection of additional material is to be expected for some species. *Bolitoglossa jugivagans* is most likely a species of the intermediate or lower elevations of the Atlantic slopes of the Cordillera Central whereas most of the *Bolitoglossa* species in western Panama are found at premontane and montane elevations (Fig. 8). The type locality lies at slightly above 1000 m, what might be classified as a transitional elevation. Thus, in comparison with the elevational preferences of other Atlantic slope *Bolitoglossa*, two different elevational distribution scenarios are imaginable for *B. jugivagans*. It might predominantly be a lowland species that reaches its upper limit at the type locality (comparable to *B. colonnea*; Fig. 8), or it might be a species of premontane elevations (comparable to *B. robusta*; Fig. 8). Members of the genus which are likely to occur in sympatry with *B. jugivagans* are *B. colonnea* and *B. robusta* and less likely, but still possible, *B. schizodactyla* and *B. anthracina* (Fig. 8).

The horizontal distribution of *Bolitoglossa jugivagans* may either be predominantly Caribbean or the species is endemic to the Fortuna area. Many species distributed along the Atlantic drainage enter the Pacific slopes in the Fortuna depression, while only few species with distributions along the Pacific drainage are present in Fortuna (Myers and Duellman 1982). Reptile and amphibian species, which are apparently endemic to the Fortuna area and vicinity, include *Anolis fortunensis* Arosemena and Ibáñez, *Sphenomorphus rarus* Myers and Donnelly, *Oedipina fortunensis* Köhler, Ponce, and Batista, and in a broader sense also *Oophaga arborea* (Myers, Daly, and Martínez). *Anolis fortunensis* and *O. fortunensis* are only known from sites on the Pacific versant of the Fortuna depression (Mayer 2011; Hertz *et al.* 2011). *Sphenomorphus rarus* is known only from the type locality on the Atlantic slopes (Köhler 2008) and *O. arborea* is found on the Atlantic slopes from the continental divide down into the lowlands, but also enters the Pacific slopes of the Fortuna depression (Myers *et al.* 1984). The type localities of the two latter species are in the immediate vicinity of the type locality of *B. jugivagans*.



**FIGURE 8.** Altitudinal distribution of *Bolitoglossa* species in western Panama. Species roughly arranged according to their occurrence on the slopes of the Cordillera Central.

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## APPENDIX

Collection Number	Species	Genbank Number
SMF 94467	<i>Bolitoglossa jugivagans</i>	KC428634
UCR 20489	<i>Bolitoglossa robinsoni</i>	JQ899161
UCR 19892	<i>Bolitoglossa aureogularis</i>	JQ899153
UCR 19893	<i>Bolitoglossa aureogularis</i>	JQ899154
UCR 19858	<i>Bolitoglossa aureogularis</i>	JQ899151
UCR 19859	<i>Bolitoglossa aureogularis</i>	JQ899152
MVZ 181260	<i>Bolitoglossa epimela</i>	AY526120
MVZ 229170	<i>Bolitoglossa gracilis</i>	AY526121
MVZ 229171	<i>Bolitoglossa gracilis</i>	AY526122
MVZ 194828	<i>Bolitoglossa subpalmata</i>	EU448107
MVZ 229172	<i>Bolitoglossa subpalmata</i>	AF416697
UCR 20483	<i>Bolitoglossa bramei</i>	JQ899159
UCR 20484	<i>Bolitoglossa bramei</i>	JQ899160
UCR 20851	<i>Bolitoglossa bramei</i>	JQ899142
UCR 20414	<i>Bolitoglossa gomezi</i>	JQ899156
UCR 20415	<i>Bolitoglossa gomezi</i>	JQ899157
UCR 20417	<i>Bolitoglossa gomezi</i>	JQ899158
UCR 20843	<i>Bolitoglossa gomezi</i>	JQ899140
UCR 20844	<i>Bolitoglossa gomezi</i>	JQ899147
UCR 20845	<i>Bolitoglossa gomezi</i>	JQ899148
UCR 20846	<i>Bolitoglossa gomezi</i>	JQ899149
UCR 20848	<i>Bolitoglossa gomezi</i>	JQ899139
UCR 20849	<i>Bolitoglossa gomezi</i>	JQ899141
UCR 20850	<i>Bolitoglossa gomezi</i>	JQ899146